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Effects of Polyethylene Bag Sizes on *In vivo* Propagation of Sugarcane (*Saccharum officinarum* L.) Planting Materials at Metahara Sugar estate of Ethiopia

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Abstract

An experiment was conducted at Metahara Sugar Estate of Ethiopia with the aim to evaluate the effects of three different polyethylene bag sizes on the propagation responses of three sugarcane genotypes in a completely randomized design with factorial treatment combination arrangements. Analysis of variance revealed that the interaction effects of polyethylene bag sizes and sugarcane genotypes have significant influence on the number of tillers per shoot, number of roots per shoot and average root length of the sugarcane genotypes studied. white colored polyethylene bag size with 10 cm diameter and 8 cm height (W10*8) gave the maximum number of tillers per shoot while white polyethylene bags with 10 cm diameter and 10 cm height (W10*10) and black polyethylene bag with 30 cm diameter and 20 cm height (B30*20) gave maximum number of roots per shoot and maximum average root length, respectively, in all the sugarcane genotypes tested. As the number of tillers per shoot is a key response variable in planting material propagation, polyethylene bag size W10*8 is found to be economical in its volume of planting media consumption and in production of maximum number of tillers per shoot of the three sugarcane genotypes tested.

Keywords:* *Polyethylene bag size; Sugarcane genotypes; Planting materials; SP70-1284; C132-81; C86-56

Introduction

Even though sugarcane is propagated through fuzz, settlings, bud chips, cane setts and micropropagation technology, the last two are the most commonly used methods for commercial propagation. One of the major items of expenditure in sugarcane cultivation is the huge planting material requirement that ranges from 22% to 25% of the total production cost [1]. Sugarcane planting material propagation via cane setts (conventional propagation method) is one of the most widely used method in commercial sugarcane propagation. However, this method has a wide range of limitations that caused the invention of micropropagation technology that has a wide range of advantages except the cost of propagation.

To minimize the cost of micropropagation, *ex-vitro* propagation/*in vivo* proliferation methods has been developed, but needs to develop complete working protocol for every step of the propagation procedure among which size of polyethylene bags used under nursery propagation is one. Polyethylene bag culture of sugarcane plants produced a significantly higher number of shoot and millable canes and a higher seed cane yields as compared with the conventional planting method, which is an advantage for seed cane multiplication [1]. In addition, with almost equal cost of cultivation, the rate of planting material multiplication using polyethylene culture method was about 35 time more than the conventional method [2].

Sizes of polyethylene bags have significant influence on plant growth under nursery conditions [3,4]. *In vivo* proliferation/*Ex vitro* propagation systems have been developed for sugarcane planting material propagation using polyethylene bag culture method. However, the appropriate polyethylene bag size for better propagation of the sugarcane plants is not optimized. Even, reports are scare on the appropriate polyethylene bag size for sugarcane plant propagation under this system. Therefore, the aim of the current study is to investigate the appropriate size of polyethylene bag size that can produce maximum number of quality tillers with low volume of planting media.

Materials and Methods

The study was conducted at Metahara Sugar Estate which is located Oromia Regional State, Ethiopia. It is situated at 8° 51, N latitude; 39° 52, N longitudes with an elevation of 950 m.a.s.l. The area has Semi-arid climatic condition with average annual rain fall of 554 mm with maximum and minimum temperature of 32.6 and 17.5, respectively. The soil type was sandy loam with a pH of 7.25, EC: 0.19 dSm^{-1}, low in available nitrogen (81.2 kg ha^{-1}) and medium in available phosphorus (11.00 kg ha^{-1}) and potassium (77.5 kg ha^{-1}). The primary acclimatized plantlets of three sugarcane genotypes were delivered from Mekelle Technology Institute Tissue Culture Laboratory and directly planted in different size polyethylene bag (white, 8 cm diameter with 10 cm height; white, 10 cm diameter with 10 cm height; black, 20 cm diameter with 30 cm height) filled with mixture of Luvisol, sand and compost in the ratio of 8:2:1. The experimental design was Completely Randomized Design with factorial treatment combination arrangements. Three sugarcane genotypes (C132-81, C86-56 and SP70-1284) with three different polyethylene bags size resulting in 3*3=9 treatment combination arrangements each replicated three time were used in the study. Each plot contains 30 pots (one plantlet a pot) and data on the number of tillers per shoot, shoot length and number of active leaves per shoot were collected from ten randomly selected plantlets on 30th day after treatment application. Then the average data were subjected to analysis of variance using SAS software version 9.2 while separation of significant means was done using REGWQ (Ryan-Einot-Gabriel- Welsch) Multiple Range Test.

Results and Discussion

Analysis of variance (ANOVA) revealed that the number of tillers per shoot, number of roots per shoot and average root length (cm) in sugarcane genotypes (SP7-1284, C132-81 and C86-56) were significantly influenced by the interaction of polyethylene bag sizes and sugarcane genotypes (polyethylene bag size*sugarcane genotypes=$p \leq 0.05$) at alpha value of 5% (TABLE 1).

All the three sugarcane genotypes showed significant variation both on the number of roots per shoot and average root length(cm) while the sugarcane genotypes SP70-1284 and C86-56 are similar in their number of tillers per shoot (TABLE 2). However, C132-81 showed the least and significantly different number of tillers per shoot than the other two genotypes studied. In sugarcane genotypes C132-81 and C86-56 gave the same number of tillers per shoot on B30*20 and W10*10 polyethylene bag sizes. C132-81 gave 2.5 tillers per shoots while C86-56 produced 4.5 tillers per shoot on both polyethylene bag sizes (TABLE 3).

TABLE 1. **Summary of ANOVA for the effects of polybag sizes and sugarcane genotype on *sugarcane planting* material propagation.**

Source of variation	DF	Mean Square		
		Number of Tillers/shoot	Number of Roots per shoot	Average Root length(cm)
Polyethylene bag size	2	3.5**	386.0***	80.91*
Genotype	2	8.0***	673.0***	26.00ns
Polybag size*Genotype	4	2.0*	98.8*	101.95**
DF=Degrees of freedom; the superscripts ns= on-significant; * = significant; ** = highly significant; *** = very highly significant at alpha=5%				

TABLE 2. **Comparison of sugarcane genotypes by their propagation responses using pooled values.**

Sugarcane genotypes	Number of tillers per shoot	Number of roots per shoot	Average root length per shoot(cm)
SP70-1284	4.67[a]	46.83a	18.68[b]
C86-56	4.67[a]	37.00[b]	17.20[c]
C132-81	2.67[c]	25.6`7[c]	20.92[a]
Remark: Means with the same letter are not significantly different			

Sugarcane genotype SP70-1284 produced different number of tillers per shoot on different polyethylene bag sizes. However, all the three sugarcane genotypes (C132-81, C86-56 and SP70-1284) gave the maximum number of tillers per shoot on W10*8 polyethylene bag size. on this polyethylene bag size (W10*8), sugarcane genotype C132-81 produced 3.0 tillers per shoot while C86-56 and SP70-1284 produced 5.0 and 6.0 tillers per shoot, respectively. In all the sugarcane genotypes tested, the lowest number of roots per shoot were obtained on B30*20 while the maximum number of roots per shoot was recorded on W10*10 polyethylene. Accordingly, the lowest number of roots per shoot of 39, 22 and 43 and the maximum number of roots per shoot of 45, 28 and 62 were obtained in sugarcane genotypes C132-81, C86-56 and SP70-1284, respectively. Similarly, maximum average root length of 26.5 cm, 21.0 cm and 29.15 cm were obtained in C132-81, C86-56 and Sp7-1284, respectively (TABLE 3). In general, polyethylene bag size W10*8 gave maximum number of tillers per shoot while W10*10 and B30*20 gave maximum number of roots per shoot and maximum average root length, respectively in all the sugarcane genotypes tested. Increase in the number of polyethylene bag size may increase the root biomass (number of roots per shoot and root length) but not increased the above ground biomass. In addition, increasing the size of polyethylene bag increase the volume of soil used and hence the cost.

TABLE 3**. Response of sugarcane genotypes to the different polyethylene bag sizes.**

Polyethylene Bag size	**Response variables**								
	Number of Tillers per Shoot			**Number of Roots per Shoot**			**Average Root Length(cm)**		
	C132-81	C86-56	SP70-1284	C132-81	C86-56	SP70-1284	C132-81	C86-56	SP70-1284
B30*20	2.50[f]	4.50[d]	5.50[b]	39[d]	22[h]	43[c]	26.5[b]	21.0[d]	29.15[a]
W10*10	2.50[f]	4.50[d]	2.50[f]	45[b]	28[f]	62[a]	10.75[h]	15.5[g]	16.40[e]
W10*8	3.0[e]	5.0[c]	6.0[a]	26.5[g]	26.5[g]	32[e]	25.5[c]	15.10[g]	16.50[e]
CV	16.30			18.23			22.46		
SE	0.52			0.47			3.11		
Remark: W30*20 = polyethylene bag size with 30 cm height and 20 cm diameter; W10*10 = polyethylene bag size with 10 cm height and 10 cm diameter; W10*8 = polyethylene bag size with 10 cm height and 8 cm diameter									

Conclusion

One of the major items of expenditure in sugarcane cultivation is the huge planting material requirement accounting for 22% to 25% of the total production cost. The conventional planting material propagation method is inefficient while micropropagation technology is costly. *Ex-vitro* propagation method was developed to increase the propagation efficiency with a relatively low cost of planting material propagation. However, this method needs for optimization at all stages of the propagation procedure mainly on the sizes of polyethylene bags used under nursery. Therefore, an experiment was conducted to evaluate the effects of three different polyethylene bag sizes on three sugarcane genotypes with factorial completely randomized design arrangements. Analysis of variance revealed that the interaction effects of polyethylene bag sizes and sugarcane genotypes have significant influence on the number of tillers per shoot, number of roots per shoot and average root length of the genotypes studied. white colored polyethylene bag size with 10 cm diameter and 8 cm height (W10*8) gave the maximum number of tillers per shoot while white polyethylene bag with 10 cm diameter and 10 cm height (W10*10) and black polyethylene bag with 30 cm diameter and 20 cm height (B30*20) gave maximum number of roots per shoot and maximum average root length, respectively, in all the sugarcane genotypes studied. Generally, as the number of tillers per shoot is a key response variable in planting material propagation, polyethylene bag size W10*8 is found to be the optimum size for *ex-vitro* propagation of the three sugarcane genotypes tested. From this result, it can be deduced that W10*8 is the optimum size to be used for *ex-vitro* propagation of the sugarcane genotypes studied. Optimization for the optimum planting media mixtures will be the future line of work to be done.

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